REMARKS

Reconsideration and allowance are respectfully requested.

Claims 1-32 are pending. New claims 31-32 are directed to the elected subject matter and should be examined in the present application.

Non-elected claims 26-30 were withdrawn from consideration by the Examiner. Applicants are grateful for the Examiner's acknowledgement that method claims 26-30 may be rejoined once the product claims are allowed. They believe, however, that the pending claims are not anticipated either by Grangette *et al.* or Pouwels *et al.* (1996), so the non-elected claims should be examined in the present application.

The specification has been amended to correct the informalities objected to by the Examiner on page 4 of the Action. A brief description of the drawing and trademark symbols are added. Withdrawal of the objections is requested.

35 U.S.C. 112 – Written Description and Enablement

Claims 16, 18-19, 23 and 25 were rejected under Section 112, first paragraph, as allegedly "failing to provide an adequate written description of the invention and failing to provide an enabling disclosure." Applicants traverse because the strain *Lactobacillus plantarum* 256 is generally available and is not subject to the deposit requirement. A person skilled in the art would have been able, as of the effective filing date, to practice the claimed inventions in accordance with Applicants' disclosure and without the need of undue experimentation.

A copy of the literature reference Johansson *et al.* (Int. J. Sys. Bacteriology 45:670-675, 1995) is attached. Johansson *et al.* discuss a variety of *Lactobacillus plantarum* strains. Table 1 of Johansson *et al.* provide a list of *Lactobacillus plantarum* strains and these include *Lactobacillus plantarum* 256. Applicants and Johansson *et al.* do not originate from the same establishment and this reference illustrates the fact that the *Lactobacillus plantarum* 256 strain was a generally available strain which the skilled person would have access to. Johansson *et al.* also provide results of restriction endo-nuclease mapping of the *Lactobacillus plantarum* strain providing further information on

the strain to allow the skilled person to check that the strain that they are employing is *Lactobacillus plantarum.*

Withdrawal of the written description and enablement rejections made under Section 112, first paragraph, is requested.

35 U.S.C. 112 – Definiteness

Claims 1-25 were rejected under Section 112, second paragraph, as being allegedly "indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention." Applicants traverse by referring to the numbered points of this section in the Action:

(a) In order to facilitate prosecution, claim 1 has been amended to delete reference to immunogenicity and hence the issue of the difference between immunity and immunogenicity has been rendered moot.

(b) Claims 2-17 have been amended to refer to "The vaccine" as requested by the Examiner.

(c) Claim 2 has been amended to specify that the vector is capable of "expressing the heterologous antigen intracellularly and/or on the cell surface." Thus, claim 2 indicates that it is the heterologous antigen that is either present inside the cell or is on the surface of the cell and not the vector.

(d) The term "such as" has been deleted from claims 3 and 15 and the preferred embodiments previously identified in these claims have been made the subject of new dependent claims 31-32.

(e) Claims 5-6 have been amended to refer to "*Vibrio cholerae*" to correct the minor spelling mistake noted.

(f) Claim 5 has been amended to refer to "*Treponema pallidum*" and "*Coxiella burnetii*" to correct the minor spelling mistakes noted.

(g) Claim 5 has been amended to refer to a "pathogenic organism" rendering the issue of whether or not *Pneumocystis pneumonia* is a microorganism moot. *Pneumocystis*

pneumonia is a pathogenic organism regardless of whether or not it is a micro-organism.

(h) Claim 5 has been amended to include the full terms for the abbreviations present in the claims and the abbreviations have been retained in brackets.

(i) Claim 7 has been amended to include the full term for the abbreviation CFA and hence now refers to a "Coli Fimbrial Antigen" with the original abbreviation being retained in brackets.

(j) As there is more than one Enteropathogenic strain of *E. coli* (EPEC) and also more than one Enteroaggregative (EaggEc) strain of *E. coli*, reference in claim 5 to strains of these particularly types of *E. coli* is not indefinite and would be understood by the skilled person.

(k) Claim 9 has been amended to specify that protective immunity is induced against the pathogenic organism from which the heterologous antigen originates. Claim 9 therefore clearly indicates what protective immunity is induced against.

(l) Claim 11 has been amended to refer to expression of the antigen "intracellularly and/or on the cell surface." The minor typographical error noted has been therefore been corrected and it is clear that the antigen is expressed inside the cell or on the surface of the cell.

(m)Claim 18 no longer refers to claim 1 and now lists all of the features of the recombinant *Lactobacillus plantarum.* Claim 18 refers to eliciting an immune response and does not refer to immunogenicity for consistency with claim 1. The scope of claim 18 is therefore self-contained and all of the features of the bacterium are apparent from the claim itself.

(n) Claim 23 has been amended to refer to "A Lactobacillus plantarum." Claim 23 is therefore consistent with claim 18, on which it depends, as both claims refer to *Lactobacillus plantarum.*

(o) The minor typographical error noted in claim 15 has been corrected and the claim now refers to "administration."

(p) The Action refers to claim 15, but from the quotes it appears that reference to claim 16 is intended. Claim 16 has been amended to specify that the *Lactobacillus plantarum* "is recombinant *Lactobacillus plantarum* 256" and hence the claim does not imply that there is more than one *Lactobacillus plantarum* 256 strain.

(q) Claims 19, 21 and 25 have been amended to provide proper antecedent basis and now refer to "The bacterium" as requested by the Examiner.

(r) Claim 21 has been amended so that it refers to "the bacterium" which has proper antecedent basis in claim 20.

(s) Claim 22 has been amended to make it clear that the immune response is induced "in" an individual.

(t) The objections raised in this point have been addressed by the amendments discussed above in points (a) to (s).

In view of the above, Applicants request withdrawal of the Section 112, second paragraph, rejection because the pending claims are clear and definite.

35 U.S.C. 102 – Novelty

A claim is anticipated only if each and every limitation as set forth in the claim is found, either expressly or inherently described, in a single prior art reference. *Verdegaal Bros. v. Union Oil Co. of Calif.*, 2 USPQ2d 1051, 1053 (Fed. Cir. 1987). The identical invention must be shown in as complete detail as is claimed. See *Richardson v. Suzuki Motor Co.*, 9 USPQ2d 1913, 1920 (Fed. Cir. 1989).

Claims 1-7 and 9-25 were rejected under Section 102(b) as allegedly anticipated by Pouwels *et al.* (J. Biotechnol. 44:183-192, 1996). Applicants traverse.

As discussed below, the reference Pouwels *et al.* (1996) (referred to hereafter as Pouwels I) does not disclose an oral vaccine comprising a *Lactobacillus plantarum* strain modified to express a heterologous antigen where the bacterium is capable of eliciting an immune response against the antigen or indeed any *Lactobacillus* bacterium capable of eliciting such a response. The subject matter of claims 1-7 and 9-25 is therefore not anticipated by Pouwels I.

Pouwels I describes various *Lactobacillus* strains. For example, at page 189, right hand column, section 3.5, Pouwels I refers to administration of *Lactobacillus* to mice and states that:

> Mice were immunized subcutaneously with two doses of 5 $x10^8$ Lactobacillus transformants expressing the HA uidA gene . . . . No systemic response to the *fusion protein was detected. Following a subsequent oral administration of Lactobacillus harbouring the HA-uidA gene* [encoding HA-β-glucuronidase] . . . . *a significant booster effect in serum was observed* [emphasis added].

Two main points can be seen from this passage:

- the administration regimen in Pouwels I is subcutaneous administration and then administration orally and hence there is no indication that the bacterium can act as an oral vaccine because the experiments referred to use a powerful subcutaneous administration step; and
- there is no indication that *Lactobacillus plantarum* was administered orally as the section only refers to administering "*Lactobacillus*" without mentioning a genus for the bacterium and it is perfectly possible that the administered bacterium was *Lactobacillus caseii*. As is well known, a generic disclosure cannot anticipate a specific element.

These points are important because there is no indication that any of the *Lactobacillus* strains discussed are able to generate an immune response against the antigen when administered orally, and indeed the strains referred to may well not do so. In Pouwels I oral administration of the bacterium is being used as a "booster" not as an oral vaccine. Oral boosting following subcutaneous administration and oral vaccination are very different things. A reference to a boosting effect following subcutaneous administration gives no indication whatsoever that a bacterium can elicit an immune response after oral administration alone.

Indeed, the experiments in Pouwels I indicate that the strain disclosed therein is unlikely to elicit an immune response. Subcutaneous administration is a very effective way to elicit an immune response and, hence, the fact that no immune response was seen in Pouwels I following subcutaneous administration points to low immunogeniccity

and the likelihood that any route of administration, let alone just oral administration, would not elicit an immune response.

It is pointed out that given one of the chief advantages of oral vaccination is that it is a quick, painless and cheap administration route, the need for a prior subcutaneous injection of antigen presumably requiring a sterile syringe or similar device would negate all of these advantages. The "boosting" effect seen in Pouwels I cannot be said to be an oral vaccination and there is no evidence that any of the strains discussed in Pouwels I could give rise to successful oral vaccination.

It is also noted that there is no disclosure in Pouwels I of oral administration of *Lactobacillus plantarum* expressing a heterologous antigen. Section 3.5 of Pouwels I, which refers to the administration of transformed bacteria expressing HA-β-glucuronidase, only refers to the administration of "*Lactobacillus*" and hence there is no disclosure that *Lactobacillus plantarum* is administered. Given that Pouwels I focuses on *L. casseii*, it appears that *L. casseii* was the *Lactobacillus* administered. There is therefore no disclosure of oral administration of transformed *Lactobacillus plantarum* expressing a heterologous antigen in Pouwels I.

It is noted that the Action also cites a number of secondary citations as evidence that the proteins referred to in Pouwels I are immunogenic. However, immunogenicity is not just a question of the proteins employed, it is very much a question of the context in which the protein is administered. A protein may give rise to an immune response when administered in a particular route or context, but fail to do so when it is administered via a different route or context. In other words, a protein may be potentially antigenic, but if it is delivered in the wrong way or using the wrong formulation no immune response may result. For example, administering a protein alone may not generate an immune response, but a protein presented on the surface of an *L. plantarum* (as in the invention) can.

Indeed, if it were that simple to generate an immune response there would not be the prior art cited there has been which illustrates various failures and how painfully slow progress, and littered with pitfalls, the development of possible vaccines can be.

As discussed below in relation to non-obviousness, the prior art highlights instances when oral administration singly failed to give rise to any immune response. One needs to select the correct administration method, type of bacterial strain, and antigen presentation in order to be successful.

Thus, it cannot be assumed that any of the strains referred to in Pouwels I would give rise to an immune response against the expressed antigen when administered orally. There is no evidence here that this is the case. Therefore, Pouwels I does not anticipate claims 1-7 and 9-25.

Claims 20-22 were rejected under Section 102(b) as allegedly anticipated by Mercenier *et al.* (Adv. Food Sci. 18:73-77, 1996). Applicants traverse.

The Action notes a minor discrepancy between claims 20 and 21 which has been addressed by amending claim 21 to refer to "the bacterium." As discussed below, the reference Mercenier *et al.* (referred to hereafter as Mercenier) does not disclose *Lactobacillus* strains where the bacterium is capable of eliciting an immune response against the heterologous antigen expressed. The subject matter of claims 20-22 is therefore not anticipated by Mercenier.

The main *Lactobacillus* strain that Mercenier focuses on is *Lactobacillus paracasei*. Section 2.3 of Mercenier refers to a non-human recombinant *Lactobacillus paracasei* strain LbTGS expressing the antigens V3 or gp41E protein. The coding sequences for the antigens are fused to sequences encoding the M6 carrier protein. Mercenier indicates at page 75, left hand column, first paragraph, that no immune response against either antigen was raised:

> None of the immunizations led to a detectable and specific anti-epitope antibody response, and no anti-CD3 or anti-gp41E APCs (antibody producing cells) could be observed by ELISPOTs.

Thus, Mercenier indicates that the strains it deals with cannot give rise to an immune response against a heterologous antigen as specified by the claims. The only immune response seen is against the M6 carrier protein, fused to the antigen, and not against the antigen.

A strain for use in an oral vaccine is not of much use if it is unable to elicit an immune response against a chosen antigen as specified by the claims! Therefore, Mercenier does not anticipate claims 22-25.

Withdrawal of the Section 102 rejections is requested because all limitations of the claimed invention are not disclosed by the cited references.

35 U.S.C. 103 – Nonobviousness

To establish a case of prima facie obviousness, all of the claim limitations must be taught or suggested by the prior art. See M.P.E.P. § 2143.03. Obviousness can only be established by combining or modifying the prior art teachings to produce the claimed invention if there is some teaching, suggestion, or motivation to do so found in either the references themselves or in the knowledge generally available to a person of ordinary skill in the art. See, e.g., *In re Fine*, 5 USPQ2d 1596, 1598 (Fed. Cir. 1988); *In re Jones*, 21 USPQ2d 1941, 1943-44 (Fed. Cir. 1992). It is well established that the mere fact that references can be combined does not render the resultant combination obvious unless the desirability of that combination is also taught or suggested by the prior art. See *In re Mills*, 16 USPQ2d 1430, 1432 (Fed. Cir. 1990). Thus, even if all elements of the claimed invention were known, this is not sufficient by itself to establish a prima facie case of obviousness without some evidence that one would have been motivated to combine those teachings in the manner proposed by the Examiner. See *Ex parte Levengood*, 28 USPQ2d 1300, 1302 (B.P.A.I. 1993). Finally, a determination of *prima facie* obviousness requires a reasonable expectation of success. See *In re Rinehart*, 189 USPQ 143, 148 (C.C.P.A. 1976).

Claims 1 and 8 were rejected under Section 103(a) as allegedly unpatentable over Pouwels *et al.* (Int. J. Food Microbiol. 41:155-167, 1998) in view of Claassen *et al.* (*Recombinant and Synthetic Vaccines*, Narosa Publ., New Delhi 407-412, 1994) which is referred to hereafter as Claassen I and Wells *et al.* (Ant. Lewuwenhoek 70:317-330, 1996). Applicants traverse.

None of the cited documents teaches or suggests an oral vaccine comprising a *Lactobacillus plantarum strain* expressing a heterologous antigen where the vaccine can elicit an immune response against a heterologous antigen as specified in claims 1 and 8. On the contrary, the cited references provide only negative results for *Lactobacillus plantarum* (administered orally) and indicates that strains with high persistence times such as *Lactobacillus plantarum* are potentially unsuitable for such a purpose.

The reference Pouwels *et al.* (1998) (referred to hereafter as Pouwels II) would not have provided one of ordinary skill in the art with any encouragement to use *Lactobacillus plantarum* in an oral vaccine because it provides only negative results for *Lactobacillus plantarum.*

One of the most notable things about Pouwels II is that it highlights the pitfalls ahead in the potential development of oral vaccines. As noted in the Action, Pouwels II states at page 162, right hand column, final paragraph:

> In initial experiments using *L. casei* and *L. plantarum* as host organisms and expression vectors . . . it was found that the levels of intracellular production were rather low (generally less than 0.2 % of total soluble protein) [emphasis added].

Pouwels II therefore hardly offers much promise that *L. plantarum* will be suitable, especially when it goes on to state in the next paragraph that whilst intraperitoneal injection with *L. casei* gave some immune response, oral immunisation with the same bacterium gave "only weak and inconsistent IgA responses in gut washes."

Pouwels II therefore indicates that both *L. casei and L. plantarum* can give poor expression with certain proteins, does not attempt oral immunisation with *L. plantarum* and shows that oral immunisation with *L. casei* was notably unsuccessful. Failure is hardly motivation for one of ordinary skill in the art to make the combination suggested in the Action.

The Action contends that one of ordinary skill in the art would have looked to the reference Wells *et al.* (referred to hereafter as Wells) because Pouwels II refers to the possible importance of persistence and the indication in Wells that *Lactobacillus plan-*

tarum persists longer than *L. casei*. However, Wells in fact teaches directly away from the use of *Lactobacillus plantarum*.

Wells cites results from a further paper by Claassen *et al.* at page 322, left hand column indicating that oral administration with *Lactobacillus plantarum* failed to give any immune response. Thus, if anything, Wells indicates that *Lactobacillus plantarum* is unsuitable for use in an oral vaccine. The reference cited in Wells is Claassen *et al.* (In: *Recombinant Vaccines: New Vaccinology* (Ed) Kurstak E, Int. Comp. Virology Org., Montreal, 1995), a copy of which is attached (referred to hereafter as Claassen II).

Claassen II discusses the use of *Lactobacillus plantarum* as a carrier to deliver antigens, in other words where antigen is artificially attached to the bacterium. Claassen II also refers to some initial experiments where proteins are expressed in *Lactobacillus plantarum* and indicates that these experiments failed. Claassen II states at page 19, final paragraph:

> no significant Ig (total) responses to β-galactosidase was obtained in ELISA after oral administration of the bacteriase when the results were compared to i.p. administration of the same bacteriae [emphasis added].

Thus, not only does Claassen II indicate that oral administration of *Lactobacillus plantarum* using galactosidase fails to bring about any significant immune response, the fact that Wells cites the results shown in Claassen II is evidence that at the time Applicants' invention was made it was generally though that *Lactobacillus plantarum* would not be suitable for use in an oral vaccine for an expressed heterologous antigen. In essence, the prior art offers no encouragement because it indicates only failure with orally administered *Lactobacillus plantarum*.

The Action contends that one of ordinary skill in the art would have used *Lactobacillus plantarum* as disclosed by Wells because Pouwels II indicates that persistence may be important. However, one of ordinary skill in the art would have given far more weight to the experiments in Claassen II (which is cited in Wells) which show that *Lactobacillus plantarum* failed to give rise an immune response following oral administration. A consideration of persistence is one thing, actual experimental data showing failure in the desired aim of oral vaccination is quite another. Furthermore, one of ordinary skill in

the art would also be aware that persistence is not necessarily a good indicator of efficacy as an oral vaccine from Wells and both Claassen I and II.

Table 1 of Wells provides persistence data for various strains of lactic acid bacteria. Table 1 indicates that the *Lactobacillus paracasei* LbTGS1.4 strain studied in Wells persists from 4 to 6 days in the gastrointestinal tract. However, despite the fact that the LbTGS1.4 strain persists at least four times longer than the *L. casei* strain in Pouwels II, the LbTGS1.4 strain still fails to give rise to an immune response. At page 323, left hand column, first paragraph, Wells states that:

> specific immune responses to an α-amylase expressed in LbTGS 1.4 were only elicited by intraperitoneal inoculation and not by a mucosal route of administration [emphasis added].

Both Claassen I and II also indicate that whether or not persistence is desirable is unclear and indeed suggest that it may in fact be a disadvantage. At page 410, lines 19 to 20, Claasen I states that:

> It is still not clear whether persistence of a live microorganism is an advantage for oral immunisation or a disadvantage. The latter might be so because of the fact that the antigenic determinant will be exposed for prolonged periods of time in the gut possibly leading to the induction of tolerance [emphasis added].

Claassen II indicates the same thing at page 19, lines 9 to 12.

It is not therefore the case that one of ordinary skill in the art would have automatically choose a strain on the basis of higher persistence given that Wells indicates that higher persistence is not an advantage for *Lactobacillus paracasei* LbTGS1.4 and both Claassen I and II indicate that it is a possible disadvantage. The prior art teaches away from the use of *Lactobacillus plantarum*.

Therefore, taken as a whole, the cited references would not have led one of ordinary skill in the art to generate an oral vaccine comprising *Lactobacillus plantarum* expressing a heterologous antigen as specified by claims 1 and 8. The prior art only provides evidence that *Lactobacillus plantarum* may be unsuitable for such a purpose and in certain circumstances can fail to elicit an immune response against an expressed

antigen when administered orally. Furthermore, both Claassen I and II indicate that the higher persistence time of *Lactobacillus plantarum* may in fact be a disadvantage.

Withdrawal of the Section 103 rejection is requested because the invention as claimed would not have been obvious to a person of ordinary skill in the art at the time it was made.

Conclusion

Having fully responded to all of the pending objections and rejections contained in this Office Action, Applicants submit that the claims are in condition for allowance and earnestly solicit an early Notice to that effect. The Examiner is invited to contact the undersigned if any further information is required.

Respectfully submitted,

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